Appl. No. : Unknown Filed : Herewith

AMENDMENTS TO THE CLAIMS

- 1. (Original) DNA encoding at least one luciferase selected from the group consisting a red-emitting luciferase and a green-emitting luciferase derived from a rail road worm and a green-emitting luciferase and an orange-emitting luciferase derived from *Rhagophthalmus ohba* stably expressed in mammalian cells, characterized in that (1) the DNA has no binding sequence for an additional transcription factor in the mammalian cells and has a codon usage for the mammal.
- 2. (Original) The DNA according to claim 1, characterized in that the mammal is human and the DNA has at least one nucleotide sequence selected from the group consisting of SEQ ID NOS:7, 10, 11 and 16.
- 3. (Original) A method for enabling the expression of DNA encoding a luciferase derived from a rail road worm or *Rhagophthalmus ohba* in mammalian cells, characterized by having
- 1) a step of altering a cDNA sequence such that no additional transcription factor is bound;
- 2) a step of changing a codon usage for insects to that for mammals in the cDNA sequence; and optionally
- 3) a step of altering the cDNA sequence with many restriction enzyme sites due to limited application at the use.
- 4. (Original) The method according to claim 3, characterized in that an amino acid sequence of the luciferase is not altered.
- 5. (Currently amended) A polypeptide which is a luciferase with a maximum luminescence wavelength of 630 nm, represented by any of the followings:
 - (1) a polypeptide having an amino acid sequence of SEQ ID NO:4; and or
- (2) a polypeptide having one or more amino acid substitutions, additions or deletions in the sequence of SEQ ID NO:4.
 - 6. (Original) The polypeptide according to claim 5, expressed in mammalian cells.

Appl. No. : Unknown Filed : Herewith

7. (Currently amended) A gene construct incorporating comprising one or two or more genes of luciferases which emit light whose wavelength does not substantially depend on a determining condition and maximum luminescence wavelength is 535 to 635 nm, to be which is stably expressible in mammalian cells.

- 8. (Currently amended) The gene construct according to claim 7 incorporating comprising 3 or more luciferase genes stably expressibly expressible in mammalian cells by incorporating wherein one or two or more genes of luciferases with a maximum luminescence wavelength of 460 to 520 nm together with one or two or more genes of luciferases which emit light whose wavelength does not substantially depend on a determining condition and maximum luminescence wavelength is 535 to 635 nm.
- 9. (Currently amended) The gene construct according to claim 7 wherein the above luciferase gene is a gene encoding at least one luciferase selected from the group consisting of a red-emitting luciferase, and a green-emitting luciferase derived from a rail road worm, and a green-emitting luciferase, and an orange-emitting luciferase derived from *Rhagophthalmus ohba* stably expressed in mammalian cells.
- 10. (Original) The gene construct according to claim 7 comprising an element for promoting efficiency of translation and/or an element for stabilizing mRNA.
- 11. (Currently amended) A gene construct capable of distinctively determining each light emitted from two or more luciferases, by incorporating comprising one or two or more genes of the luciferases which emit light whose wavelength does not substantially depend on a determining condition and if necessary a gene of the luciferase which emits light whose wavelength is different and does not substantially depend on the determining condition under the control of different promoters.
- 12. (Currently amended) An expression vector containing the gene construct according to claim 7 any of claims 7 to 11.

Appl. No. : Unknown Filed : Herewith

13. (Currently amended) Mammalian cells transformed with the gene construct according to <u>claim7</u> any of claims 7 to 11 or the expression vector according to <u>claim 12</u>.

- 14. (Currently amended) Mammalian cells stably expressibly incorporating comprising two or more stably expressing genes of luciferases which emit mutually distinct light whose luminescence wavelength does not substantially depend on a determining condition under the control of different promoters in the mammalian cells.
- 15. (Currently amended) The mammalian cells according to claim 13 or 14 wherein two or more of the above luciferases have a maximum luminescence wavelength of 535 to 635 nm and can emit with one substrate.
- 16. (Original) The mammalian cells according to claim 15 comprising a red-emitting luciferase gene from a rail road worm and further comprising at least two or more selected from the group consisting of a green-emitting luciferase gene from the rail road worm, a green-emitting luciferase gene from *Rhagophthalmus ohba*, an orange-emitting luciferase from *Rhagophthalmus ohba*, and a blue-emitting luciferase gene under the control of different promoters.
- 17. (Currently amended) The mammalian cells according to claim 14 stably expressibly incorporating comprising genes of three or more luciferases which emit mutually distinct light whose luminescence wavelength does not substantially depend on a determining condition under the control of different promoters in the mammalian cells.
- 18. (Currently amended) The mammalian cells according to claim 14 having comprising three or more luciferase genes under the control of different promoters wherein a first luciferase gene is under the control of a constantly expressed promoter, a second luciferase gene is under the control of a toxicity assessing promoter, and remaining one or more luciferase genes are under the control of a promoter subjected to assessment.
- 19. (Currently amended) The mammalian cells according to claim 14 having comprising three or more luciferase genes under the control of different promoters wherein a first

Appl. No.

Unknown

Filed

Herewith

luciferase gene is under the control of a constantly expressed promoter, a second luciferase gene is under the control of a pseudopromoter, and remaining one or more luciferase genes are under the control of a promoter subjected to assessment.

- 20. (Currently amended) The mammalian cells according to claim 14 having comprising 4 or more luciferase genes under the control of different promoters, wherein a first luciferase gene is under the control of a constantly expressed promoter, a second luciferase gene is under the control of a toxicity assessing promoter, a third luciferase gene is under the control of a promoter of a protein which accepts an external factor, and remaining one or more luciferase genes are under the control of a promoter subjected to assessment.
- 21. (Currently amended) The mammalian cells according to claim 14 having comprising 4 or more luciferase genes under the control of different promoters, wherein a first luciferase gene is under the control of a constantly expressed promoter, a second luciferase gene is under the control of a pseudopromoter, a third luciferase gene is under the control of a promoter of a protein which accepts an exogenous factor, and remaining one or more luciferase genes are under the control of a promoter subjected to assessment.
- 22. (Currently amended) The mammalian cells according to claim 14 having comprising two luciferase genes under the control of different promoters, wherein a first luciferase gene is under the control of a constantly expressed promoter, and a second luciferase gene is under the control of a toxicity assessing promoter.
- 23. (Currently amended) The mammalian cells according to claim 14 having comprising two luciferase genes under the control of different promoters, wherein a first luciferase gene is under the control of a constantly expressed promoter, and a second luciferase gene is under the control of a pseudopromoter.
- 24. (Currently amended) A method for screening drugs including comprising a step of culturing the mammalian cells according to claim 18 any of claims 18 to 21 in the presence of a drug candidate compound in a medium of the mammalian cells, a step of quantifying an amount of the above luciferase in the presence or absence of the candidate compound, and a step

Appl. No.

Unknown

Filed

Herewith

of assessing an effect of the candidate compound on a promoter subjected to assessment, which is linked to at least one luciferase.

- 25. (Currently amended) A system for multiply determining transcription activity of each promoter linked to each luciferase before and after a change of a culture environment by changing the culture environment of the mammalian cells according to claim 13 any of claims 13 to 23, and assessing expressed amounts of two or more luciferases which emit mutually distinct light whose luminescence wavelength does not depend on a determining condition.
- 26. (Currently amended) The system according to claim 23 <u>capable of for</u> simultaneously determining expressed amounts of two or more luciferases.
- 27. (Original) The system according to claim 23 capable of determining expressed amounts of three or more luciferases.